

Interpretive Data:

Background Information for Alpha Globin (*HBA1* and *HBA2*) Sequencing and Deletion/Duplication

Characteristics:

Alpha thalassemia is caused by decreased or absent synthesis of the hemoglobin alpha chain resulting in variable clinical presentations. Alpha (+) thalassemia results from variants of a single *HBA2* globin gene (-a/aa) and is clinically asymptomatic (silent carrier). Alpha (0) thalassemia (trait) is caused by variants of both *HBA2* globin genes (-a/-a) or variants in the *HBA1* and *HBA2* globin genes on the same chromosome (--/aa) and results in mild microcytic anemia. Hemoglobin H disease occurs due to variants of three alpha globin genes (--/-a) and results in hemolysis with Heinz bodies, moderate anemia, and splenomegaly. Hb Bart Hydrops Fetalis Syndrome results when variants occur in all four alpha globin genes (--/--) and is lethal in the fetal or early neonatal period. Alpha globin gene triplications result in three active alpha globin genes on a single chromosome. Nondeletional alpha globin variants may be pathogenic or benign; both may result in an abnormal protein detectable by hemoglobin evaluation. Pathogenic nondeletional variants often have a more severe effect than single gene deletions.

Incidence: Carrier frequency in Mediterranean (1:30-50), Middle Eastern, Southeast Asian (1:20), African, African American (1:3).

Inheritance: Autosomal recessive.

Cause: Pathogenic variants in the alpha globin gene cluster.

Clinical Sensitivity: 99 percent.

Methodology: Bidirectional sequencing of the *HBA1* and *HBA2* coding regions, intron-exon boundaries and 3' polyadenylation signal. Multiplex ligation-dependent probe amplification (MLPA) of the alpha globin gene cluster (*HBZ*, *HBM*, *HBA1*, *HBA2*, *HBQ1*) and its HS-40 regulatory region.

Analytical Sensitivity and Specificity: 99 percent.

Limitations: Diagnostic errors can occur due to rare sequence variations. Sequence analysis will not detect all regulatory region variants or variants in alpha globin cluster genes other than *HBA1* and *HBA2*. Sequencing of both *HBA1* and *HBA2* may not be possible in individuals harboring large alpha globin deletions on both alleles. This assay is unable to sequence *HBA2-HBA1* fusion genes; thus, *HBA1* or *HBA2* sequence variants occurring in cis with a 3.7 kb deletion or other *HBA2-HBA1* hybrid gene will not be detected (e.g., HbG Philadelphia will not be detected when in cis with the 3.7 kb deletion). It may not be possible to determine phase of identified sequence variants. Specific breakpoints of large deletions/duplications will not be determined; therefore, it may not be possible to distinguish variants of similar size. Individuals carrying both a deletion and duplication within the alpha globin gene cluster may appear to have a normal number of alpha globin gene copies. Rare syndromic or acquired forms of alpha thalassemia associated with *ATRX* variants will not be detected.

This test was developed and its performance characteristics determined by ARUP Laboratories. It has not been cleared or approved by the U.S. Food and Drug Administration. This test was performed in a CLIA-certified laboratory and is intended for clinical purposes.

Counseling and informed consent are recommended for genetic testing. Consent forms are available online.